Erythropoietin and Anemia

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Recombinant human erythropoietin (rHuEPO) has revolutionized the treatment of anemia of chronic renal failure. RHuEPO has been shown to increase survival, decrease hospitalizations, improve brain and cognitive function, and improve quality of life for renal patients. Much has been learned about the normal and pathologic physiology of anemia because rHuEPO has become available to investigators, and this has been widely applied. Additional work is needed in better defining the sites of production of endogenous EPO as well as the nature and control of the oxygen sensor(s) in the kidney. Remaining clinical issues related to this remarkable compound include predicting and overcoming resistance; avoiding iron deficiency; determining the appropriate target hemoglobin; increasing the use strategies such as subcutaneous administration to increase efficiency; and devising a more rational payment scheme.

ANEMIA IS ONE of the most common complications of chronic renal disease, described by Richard Bright nearly 2 centuries ago.[1] Anemia first appears when the glomerular filtration rate falls below 40 mL/min, and is present in the majority of patients who have end-stage renal disease (ESRD) and require renal replacement therapy. When untreated, anemia causes or contributes to weakness, fatigue, insomnia, depression, cognitive dysfunction, decreased libido, and left ventricular hypertrophy. Although there may be contributing factors to the anemia, or mitigators of the severity of anemia in some patients, the primary cause of the anemia is the lack of sufficient quantities of endogenous erythropoietin (EPO).

Before 1989, the year recombinant human erythropoietin (rHuEPO) was approved for use in the United States by the Food and Drug Administration (FDA), anemia in ESRD patients was often severe, and treatment with regular blood transfusions and administration of androgenic steroids was required, often with significant resultant side effects and complications. Even with these therapies most patients remained severely anemic, with hemoglobin values generally 10 g/dL or less.

The term "erythropoietin" was not used until nearly 100 years after Bright's recognition of renal anemia, and in 1953 Erslev and colleagues confirmed the presence of this substance in the blood of anemic animals.[2] Subsequent studies by Goldwasser and colleagues showed that the kidneys were the source of this hormone, which was produced in response to a fall in oxygen delivery to the renal parenchyma. The isolation and cloning of the *EPO* gene by Lin and colleagues was the breakthrough that led to the current rHuEPO, as well as provided large quantities of this remarkable substance for use by investigators.[3]

The availability of rHuEPO is certainly one of the milestones in nephrology in the past 20 years. In addition to dramatically improving the clinical outcomes and quality of life for ESRD patients, it has provided a key tool for investigators to better understand the normal and abnormal physiology associated with anemia in a variety of disease states. It is through such basic and translational research that scientific knowledge in general and in biomedicine advances, and can be applied to the care of patients. This review will touch on the highlights of our current knowledge in this field.

PHYSIOLOGY OF EPO

Structure of Endogenous EPO

The gene for EPO was isolated in 1985.[3] It is located on chromosome 7 and consists of 5 exons and 4 introns. EPO is a 166 amino acid peptide that has 2 sulfide bridges, 4 sites of carbohydrate attachment, and a molecular weight of 30,000 daltons.[4] To be secreted, and to have significant biological activity, the protein molecule must be glycosylated. This is accomplished with 4 complex carbohydrate chains containing sialic acids.[5] It is the sialic acid moieties that allow EPO to circulate in the blood for a sufficient time to reach the bone marrow and attach to EPO receptors.[6] Modification of the N-linked sugar chains increases the uptake

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EXHIBIT 1

by the kidney, thus decreasing the plasma level of the hormone, and increasing its plasma clearance.[7]

Physiology of Erythropoiesis

Synthesis and Stimulus

EPO is synthesized by fibroblast-derived interstitial cells found near the base of the proximal tubular cells in the cortex and outer medulla of the kidney.[8,9] Because expression of EPO in vitro in isolated renal cell lines has not yet been accomplished, it is difficult to study the cellular mechanisms that control its production in the kidney. Normal plasma levels of EPO are 8 to 24 mU/mL which requires daily continuous synthesis of 2 to 3 U/kg body weight.[10] The production of EPO is regulated by oxygen availability and increases when decreased arterial pO_2, anemia or increased hemoglobin affinity is detected.[11] It is known that activation of EPO gene expression requires specific transcription factors including hypoxia-inducible factor-1, a DNA binding transcriptional complex that is essential for enhancer function of this gene.[12] The adult liver also synthesizes EPO, although to a lesser degree than the kidney,[13] with production taking place in the hepatocyte[14,15] and the Ito cell.[16]

In addition to the kidney and liver, *EPO* gene expression has been found in the lung, spleen,[17,18] brain, testes, and ovaries.[19] Although it is known that EPO regulates the growth and development of erythroid progenitor cells, other functions may exist given the fact that functional EPO receptors have been found on human, rat, and mouse kidney cells.[20]

Life Cycle of Erythropoiesis

The development of an erythrocyte begins with the differentiation of a pool of pluripotent stem cells from a stochastic differentiation of bipotential or multipotential progenitors.[21] These committed progenitor cells are stimulated from the G0 to G1 cell cycle stage by interleukin (IL) IL-1 and 6, and granulocyte colony-stimulating factor (G-CSF).[22] From G1, the progenitor cells differentiate into the burst forming unit-erythroid (BFU-E), stimulated by IL-3 and GM-CSF. The BFU-E is different from the previous cell because it has EPO receptors but loses the ability of self-renewal. The BFU-E and the Colony Forming Unit-Erythroid (CFU-E) cannot be identified by specific morphologic features. The CFU-E is more sensitive and dependent on EPO for survival and differentiation into a erythroblast. The number of CFU-E is proportional to the EPO level.[23,24] With continued maturation, the CFU-E will become activated and develop into an erythroblast[25] which is unaffected by EPO, and then continues to develop into a pronormoblast and then an erythrocyte.

Erythropoiesis is influenced by cytokines and growth factors other than EPO. IL-9 and granulocyte-macrophage CSF (GM-CSF) have been shown to have burst promoting effects whereas IL-3 causes BFU-E to propagate.[26,27] In addition, a subset of BFU-E, thought to be less mature, is able to survive without EPO if IL-3 or GM-CSF is present.[28] Insulin or insulin-like growth factor-1 (IGF-1) is also needed for optimal transformation of a CFU-E to an erythroblast.[29] Steel factor (SF, also called c-kit ligand) has marked synergistic activity on BFU-E cultured in the presence of EPO[30-32] and is necessary for the normal development of CFU-E.[33] EPO, along with either Steel factor, IL-3, or GM-CSF are required by the BFU-E to increase the number of cells and mature to form CFU-E.[34] Other factors such as androgens, thyroxine, somatomedin, and catecholamines seem to increase the growth of CFU-E but are not essential.[35] Vitamin A has been shown to increase the concentration of EPO when added to hepatoma cell lines in medium.[36] Cytokines that have a negative effect on erythropoiesis include IL-1α, and β, IL-2, tumor necrosis factor α (TNF-α), and transforming growth factor-β (TGF-β).[37-39]

EPO has been found to have effects after a CFU-E becomes an erythroblast. EPO appears to be involved in preventing neocytolysis, which is a process of selective hemolysis of the youngest circulating erythrocytes when there is an overabundance of red blood cells.[40] Neocytolysis has been documented to occur in astronauts and individuals who live at high altitude and descend to sea level.[41-43] The neocytolysis has been prevented with low doses of subcutaneous EPO.[43] Neocytolysis has been shown to contribute to the anemia of renal disease, when studied in hemodialysis patients.[44]

The EPO receptor is a 55,000 Dalton transmembrane protein.[45] This protein is a 507 amino acid polypeptide having a single membrane-spanning domain, with the extracellular N-terminal region containing the EPO-binding domain and the C-

terminal intracellular region participating in signal transduction.[46] The signal transduction acts by activating tyrosine kinase that phosphorylates a set of intracellular proteins resulting in the release of second messengers.[47] It is unclear how these second messengers act but it is thought that the signals prevent apoptosis of the progenitor cells.[48]

RHuEPO

Available Forms

Currently there are 2 forms of rHuEPO in wide clinical use worldwide and an additional form, epoetin omega currently under evaluation (Bren AF, personal communication, September 2000). In addition, gene-activated EPO has been undergoing clinical evaluation as well. At the present time, however, Epoetin alfa is the only form available in the United States, whereas Epoetin beta with a similar efficacy and safety profile, is available in other countries. Because of the need for glycosylation, Epoetin alfa is manufactured in mammalian (Chinese hamster ovary) cells.

Pharmacology of rHuEPO

The metabolism of EPO does not appear to depend on the kidney or liver. The half-life of intravenous rHuEPO is 4 to 9 hours and greater than 24 hours when given subcutaneously.[49] Animal studies have shown that removal of the kidneys or liver does not affect the volume of distribution, mean residence time, or half-life of 125-labeled rHuEPO.[50] EPO is thought to be metabolized by a compartment known as the "erythron" which is the total mass of cells in the erythropoietic pathway that is EPO dependent.[51] The distribution volume is similar in uremic and normal individuals after intravenous dosing.[52] The termination elimination half-life, however, is longer, and the whole body clearance is reduced, in uremic patients. The bioavailability of rHuEPO after subcutaneous dosing is lower in uremic patients when compared with normal individuals, with lower and delayed peak concentration. The pharmacokinetics of rHuEPO are nonlinear.[53,54]

Route of Administration

RHuEPO can be given intravenously, subcutaneously, and intraperitoneally. The intraperitoneal administration of rHuEPO is generally not recommended because of low bioavailability (2% to 12%) when compared with intravenous and subcutaneous dosing.[55,56] If intraperitoneal rHuEPO must be used, it should be administered when there is no dialysate in the peritoneal cavity.[57] Subcutaneous injection frequently gives a plasma level that is 10% of what is seen with intravenous dosing. The time to peak concentration after subcutaneous administration is usually greater than 10 hours, whereas the bioavailability has a large range from 16% to 50%.[58-63] Bioavailability has also been shown to be different depending on where the subcutaneous injection is given, with greater bioavailability when rHuEPO is given in the thigh compared with the abdomen or arm.[64] The amount of skin fold thickness of the patient has been shown to inversely influence the effectiveness of a given dose of subcutaneous rHuEPO.[65]

Subcutaneous administration of rHuEPO has many advantages compared with intravenous administration.[66] Pharmacodynamics are more physiologic with subcutaneous dosing versus intravenous dosing.[53,67-70] The dose of rHuEPO can be lowered by 25% to 50% when using subcutaneous dosing as compared with intravenous dosing,[53,64,68-71] resulting in a substantial cost saving. Subcutaneous dosing will allow for rHuEPO levels to be sustained above basal levels throughout the week in hemodialysis patients,[53] minimizing wide fluctuations in EPO-dependent apoptosis and resulting in more efficient erythropoiesis.[51] If the frequency of dosing with subcutaneous rHuEPO is decreased from thrice weekly, some of these advantages may be lost.[53] Smaller, daily doses of rHuEPO, on the other hand, have been shown to be effective in significantly reducing the total weekly rHuEPO dose.[72,73]

Although subcutaneous rHuEPO has been shown to be more effective that intravenous rHuEPO, this is not the method of dosing most frequently used in the United States and many other parts of the world. Subcutaneous rHuEPO from single-dose vials causes more patient discomfort than intravenous rHuEPO. In addition, the need to perform needle sticks repetitively can also be of concern to the health care providers who actually administer the drug. There are a number of strategies that can be used to encourage the use of subcutaneous dosing.[66] These include using the multidose vial of Epoetin alfa which contains the preservative benzyl alcohol that acts as a local

anesthetic; using a smaller gauge needle for injection (eg, 29 gauge); encouraging the patient to do their own injections; using a small injection volume; rotating the injection site; and providing education on the value of the subcutaneous route to patients and care-givers.

Dosing of rHuEPO

RHuEPO, when given subcutaneously to adult ESRD patients, is generally initiated at 80 to 120 units/kg/week, given in 2 to 3 doses. Pediatric patients <5 years of age commonly require higher doses (300 units/kg/week) than older pediatric patients[74,75] or adults. The initial intravenous dose of rHuEPO should be 120 to 180 units/kg/week given in 3 divided doses.[66]

The dose that is initially selected should achieve the target hemoglobin/hematocrit (Hgb/Hct) within a 2- to 4-month period through a slow increase of the Hgb/Hct. During the initial dosing of rHuEPO and with dose increases or decreases, a Hgb/Hct should be measured every 1 to 2 weeks until a stable target Hgb/Hct is achieved. Frequent monitoring initially is needed to detect a poor or overly rapid response to rHuEPO that would require a dose adjustment. After a stable rHuEPO dose and target Hgb/Hct are achieved, Hgb/Hct should be monitored every 2 to 4 weeks.[66] The maintenance dose varies tremendously among patients from 1,000 units to over 10,000 units per treatment to maintain a Hgb between 11 and 12 g/dL,[53-55] although the average dose in the United States currently is around 17,000 units/week (Collins A, personal communication, September, 2000). In general, doses of rHuEPO should be changed, when necessary, by no more than 25%, and withdrawing rHuEPO to permit a fall in Hgb that is higher than the upper target limit is rarely necessary and should be discouraged. Such dosage modifications are generally not necessary or effective more frequently than every 2 to 4 weeks.[66]

REFRACTORINESS TO rHuEPO

A dose of 450 units/kg/week intravenously or 300 units/kg/week subcutaneously will achieve target Hgb/Hct in 96% of patients within 4 to 6 months as long as there are adequate iron stores.[76,77] However, rHuEPO resistance should be considered to be present when there is an unsuccessful attempt to achieve a target Hgb/Hct, in a patient with sufficient iron stores, within 4 to 6 months or failure to maintain the Hgb/Hct at the previously effective dose. Iron deficiency is the most common cause of initial or acquired resistance to rHuEPO, and is still highly prevalent, despite the availability of parenteral iron compounds.[78,79] Absolute iron deficiency is present in patients with chronic renal failure when the ferritin <100 ng/mL and/or the transferrin saturation <20%. Functional iron deficiency is present when the demands for iron exceed its availability. In this situation, the serum ferritin >100 ng/mL and the transferrin saturation is usually >20%, indicating the absence of absolute iron deficiency.[80] Iron deficiency is common in hemodialysis patients because of the chronic blood loss that occurs from laboratory tests and blood remaining in the dialyzer and tubing, as much as 4,500 mL on an annual basis. In addition, rHuEPO, by accelerating erythropoiesis, further increases the demand for iron. In hemodialysis patients, oral iron is infrequently sufficient to meet iron needs, and the best way to replete iron stores in these patients is with intravenous iron. Patients on peritoneal dialysis and predialysis patients may be able to maintain iron stores with oral iron, although GI side effects may limit the effectiveness of this therapy.[81] In addition, as transferrin saturation rises above 15%, intestinal iron absorption becomes impaired, thus decreasing the bioavailability of oral iron.[82]

Inflammatory states are also frequently the cause of poor response to rHuEPO. These may result from chronic infections, postsurgery, rheumatologic diseases, or the dialysis process itself, particularly if bioincompatible dialysis membranes or pyrogen-containing water are used. The inflammatory state is usually associated with inflammatory cytokines such as IL-1-β, and TNF-α that decrease bone marrow responsiveness to rHuEPO.[83,84] In addition, in the presence of inflammation there is an impairment of iron release from the reticuloendothelial system ("reticuloendothelial blockade").[85] An elevated C-reactive protein may be a useful diagnostic test when chronic inflammation is suspected. It has been shown to be a strong predictor of rHuEPO resistance in hemodialysis and peritoneal dialysis patients.[86-87]

The ability of angiotensin-converting enzyme inhibitors (ACEI) to decrease rHuEPO responsiveness has been debated. EPO production has been shown to decrease in chronic renal failure patients treated with ACEI.[88] In addition, postrenal trans-

plant erythrocytosis has been effectively treated with ACEI.[89] Activation of the renin-angiotensin system is thought to increase endogenous EPO production in peritubular fibroblasts.[90] The exact mechanism of how ACEI may decrease erythropoiesis in patients receiving exogenous EPO is unknown. Some hypotheses include a reduction in production of IL-12, which is known to stimulate erythropoiesis,[91] and a decrease in angiotensin II-stimulated erythroid progenitor cell maturation, an effect that has been shown in vitro.[92] The data are conflicting as to whether ACEI inhibit erythropoiesis in dialysis patients receiving rHuEPO because many of the studies were uncontrolled or had small numbers of patients.[93-96] Thus, it may be reasonable to decrease the dose of ACEI or stop the drug if the patient is showing resistance to rHuEPO, and no other causes can be found.

Blood loss through the GI tract, repeated episodes of dialyzer clotting, or hemolysis should also be considered in the evaluation of rHuEPO resistance. Recently, hemolysis of erythrocytes has been shown in hemodialysis patients because of high levels of chloramine present in the water used to prepare dialysate.[97-99]

L-carnitine deficiency has been suggested by some to contribute to refractoriness to rHuEPO. Dialysis patients may have decreased predialytic serum concentrations of free L-carnitine, and low muscle carnitine content.[100-102] The benefits of L-carnitine supplementation in patients refractory to rHuEPO are still unclear. Some studies have shown that L-carnitine may increase reticulocyte count[103] or improve mechanical stability of erythrocytes.[104] Unfortunately, however, the majority of studies that have evaluated the benefit of L-carnitine in this setting have yielded conflicting results.[105-108]

Hyperparathyroidism, if severe and causing osteitis fibrosa cystica with bone marrow fibrosis, has also been shown to cause rHuEPO resistance.[109] Other explanations for the relationship between parathyroid hormone (PTH) and rHuEPO resistance include a direct toxic effect of PTH on erythroid precursors, PTH-induced hemolysis, or PTH inhibition of endogenous EPO production.[110-111] Although there is little in vivo evidence that any of these mechanisms is active, other than bone marrow fibrosis, it has been shown that medical or surgical treatment parathyroidectomy is effective in reducing rHuEPO resistance.[112-116]

CLINICAL EFFECTS OF PARTIAL ANEMIA CORRECTION

Quality of Life

The anemia of renal disease may be manifested by a variety of symptoms, differing from patient to patient. Symptoms related to quality of life include alterations in psychosocial function, poor physical and mental function scores on quality of life instruments, sexual dysfunction, and a high incidence of depression. Improvements in a variety of these symptoms after partial correction of anemia with rHuEPO has been shown using the KDQOL-SF 36, the Karnofsky index, the Sickness Impact Profile, and many other indexes.[117-121] More complete correction of anemia, to a Hgb of greater than 12 g/dL, in a cross sectional study, showed an even greater improvement in quality of life on the Sickness Impact Profile.[122] In addition, an improvement in sexual function occurred, which may be secondary to an increased serum testosterone or decreased lutenizing hormone.[123] Exercise capacity improves significantly in patients treated with rHuEPO, including those patients with coronary artery disease and rest angina.[124-125] Another indirect way that rHuEPO can lead to improved quality of life is by making renal transplantation more likely by avoiding the sensitization associated with multiple blood transfusions.[126] Quality of life has been shown to improve significantly after transplantation.[127]

Brain and Cognitive Function

Despite adequate dialysis, ESRD patients still have mild neurobehavioral impairments, with abnormalities persisting on cognitive function and electrophysiologic testing.[128-130] Although there are many contributing factors to these abnormalities, it is clear that anemia is one of the important factors affecting brain function in these patients,[131] perhaps because of alterations in cerebral oxygen delivery in the presence of anemia.[132] Cognitive function in these patients improves when the Hct is increased to 36%.[133-136] Normalizing the Hct to 40% to 45% may lead to even further improvement in neurocognitive function, shown using quantitative electroencephalograpic techniques.[137]

Cardiovascular Disease

Cardiovascular disease still accounts for over 50% of total deaths in patients on dialysis,[138] with

significant cardiac disease often present in ESRD patients before starting on dialysis.[139] The anemia of renal failure causes hypoxic vasodilatation, increased sympathetic stimulation, and decreased blood viscosity.[140] The latter exacerbates peripheral vasodilatation and contributes to decreased total systemic vascular resistance.[141] Cardiac output increases in a compensatory fashion to maintain adequate perfusion to tissues.[142] Serial echocardiographic studies in ESRD patients after the initiation of chronic dialysis have shown that left ventricular dilatation with compensatory hypertrophy is the major pattern of disease progression.[143] Patients with left ventricular dilatation and normal systolic function have a poor prognosis, with a high mortality rate at 2 years. The prognosis in patients with left ventricular hypertrophy with normal systolic function, on the other hand, is better.[144] Anemia is an important risk factor for these cardiac abnormalities, and partial correction of anemia with rHuEPO leads to regression of left ventricular hypertrophy in most patients. Patients with anemia of renal failure have a higher risk of development of cardiomyopathy that increases the risk of developing congestive heart failure, thus being a predictor of mortality in ESRD patients.[145]

In numerous studies, partial correction of anemia with rHuEPO was shown to decrease hypoxic vasodilatation, increase systemic vascular resistance, and reduce cardiac output.[146-153] In addition, partial correction of anemia with rHuEPO may result in decreased left ventricular mass and volume.[146-148,150-156] It is less clear, however, whether these changes will result in improved mortality or regression of cardiomyopathy. The Canadian randomized controlled trial of Hgb normalization with rHuEPO showed that there was no significant regression of left ventricular mass index in patients with left ventricular hypertrophy, but patients with left ventricular dilatation had slowed progression.[157] The Amgen Normal Hematocrit Cardiac Trial studied hemodialysis patients with coronary artery disease and/or symptomatic heart failure and found that the group of patients who were randomized to the normal Hct had a 30% increased risk of death or myocardial infarction when compared with the control group,[158] although the study was stopped before these group differences reached statistical significance. The effect of normalizing Hgb with rHuEPO on cardiomyopathy progression was recently examined in hemodialysis patients with concentric left ventricular hypertrophy or left ventricular dilatation.[159] Study results showed that normalizing the Hgb did not cause regression of the concentric left ventricular hypertrophy or left ventricular dilatation.

Mortality

Several epidemiologic studies have shown the survival benefits of partial correction of anemia with rHuEPO. Madore et al retrospectively studied 21,899 patients who were on hemodialysis throughout the United States from October 1 through December 31, 1992.[160] Compared with patients with a Hgb concentration of 10.0 to 11.0 g/dL, those with a Hgb concentration ≤8.0 g/dL had a 2-fold increase in the odds ratio of death. There was no decrease in the odds ratio of death with Hgb >11.0 g/dL. However, other patient characteristics and laboratory data were not adjusted for in this analysis. Ma et al retrospectively studied Medicare patients on hemodialysis and included in the analysis comorbidity ("severity of disease") adjustment, and data on hospitalizations.[161] The group of patients with Hct levels less than 30% had an overall relative risk of death that was 12% to 33% higher than patients with Hct levels in the range of 30% to less than 33%, even after adjusting for severity of disease. In an additional report using USRDS data, Collins et al evaluated whether changes in Hct level, rather than just a single or average value, over a 6-month period, would affect mortality rates.[162-164] As had been shown in the previous studies, patients with the lowest Hcts had the highest risk of death. Lower Hct levels were more likely for those patients who were younger, female, and African American, as well as those with more comorbidities, hospital days, blood transfusions, and vascular access procedures. Patients that started with a low Hct that then rose had a risk of death that was equivalent to the group whose Hct had been stable at the higher level for the full year. This study showed that a patient with a low Hct that can be improved with rHuEPO has a good prognosis, whereas refractoriness to rHuEPO in a patient with a low Hct is a poor prognostic sign.

Hospitalizations

There are only a small number of studies, which have assessed the relationship between the level of anemia and hospitalizations in rHuEPO-treated pa-

tients. In a case-controlled study by Churchill et al, patients treated with rHuEPO had fewer hospitalizations and decreased length of stay compared with a matched group of patients who did not receive rHuEPO, but the differences were not statistically significant.[165] Xia et al analyzed the data from the mortality analysis of Ma et al and found that patients with Hct levels <30% had a 14% to 30% increased risk of hospitalization without disease severity adjustment which fell somewhat to 7% to 18% with disease severity adjustment.[166] Patients with Hct levels in the 33% to 36% range had the lowest risk for hospitalization.

SIDE EFFECTS OF rHuEPO

Hypertension

RHuEPO is well known to be associated with the development of or worsening of preexisting hypertension within weeks to months of initiation in approximately 25% of treated patients.[167-170] Of interest, however, is a recent study of the effects of normalizing the Hgb on ambulatory blood pressure in patients with cardiac disease, which did not confirm this.[171] The specific mechanism of rHuEPO-related hypertension is unknown, but there are many theories based on animal and human studies, most focused on the possible role of stimulation of endothelin by rHuEPO.[172-175] In fact, one study showed that endothelin antagonists were able to inhibit rHuEPO-induced hypertension.[176] It has also been shown that EPO induces nitric oxide synthase activity without affecting endothelin-1 release, suggesting that the mechanism of rHuEPO-related hypertension is not a direct effect on endothelial cells.[177] In contrast, animal studies have suggested that rHuEPO-related hypertension may be caused by resistance to the vasodilatory action of nitric oxide.[178-179] An increase in calcium uptake has also been shown in vitro with endothelial cells treated with rHuEPO.[180-181] What is clear is that that the increase in Hgb and Hct are not themselves responsible for the rise in blood pressure.[176-179,182-183] RHuEPO-induced hypertension can easily be treated by initiating or increasing antihypertensive medications or increasing the amount of ultrafiltration during dialysis. Holding or decreasing the dose of rHuEPO has not been shown to be effective or necessary, although this may be tried if the hypertension is refractory to treatment.

Access Clotting/Other Side Effects

Although initial concern existed about the possibility of a variety of other side effects when rHuEPO is given, none of these has proven to be clinically manifested at the currently achieved target Hgb levels. Seizures were initially thought to occur with a higher frequency in patients receiving rHuEPO,[76] but a controlled study did not show this correlation.[184] In addition, an increase in vascular access thrombosis has not been convincingly shown.[185] On the other hand, the Amgen Normal Hematocrit Cardiac Trial did show an increased risk of vascular access thrombosis in the high Hct group (39% *v* 29%),[158] in native fistulas as well as grafts, suggesting that this issue bears further scrutiny. Hyperkalemia was also initially observed with early use of rHuEPO,[186] but more recent data suggest that significant hyperkalemia is not a serious rHuEPO-related problem.[187-190] Of more concern is a recent report that EPO stimulates proliferation of human renal cell carcinoma cells in vitro.[191] However, to date there is no indication that renal cell carcinoma has been occurring more frequently since the widespread use of rHuEPO developed.

TARGET HEMOGLOBIN

There is continued debate about the appropriate target Hgb in renal patients treated with rHuEPO. When the planning for the phase III trial for rHuEPO was undertaken, hematologists proposed a normal Hgb target whereas nephrologists recommended a lower level because of concern about possible vascular complications of higher Hgbs.[76] A compromise was reached and a target Hct of 32% to 38% was used. Subsequently, the National Kidney Foundation Dialysis Quality Initiative (NKF-DOQI) guidelines recommended a target Hgb of 11 g/dL to Hgb 12 g/dL, and this is supported by a large body of evidence.[66] In fact, a number of nephrologists consider that an even lower Hgb may be adequate to achieve maximum clinical benefits.[192]

In contrast, however, is a growing body of evidence that suggests that normalization of Hgb may be beneficial in some patients. The greatest amount of data in this regard relate to brain function, with clear evidence that brain electrophysiology improves when Hgb is normalized compared with when Hgb levels are lower.[137] Recent studies have confirmed that brain circulation, metabolism, and

oxygen delivery in dialysis patients occurs at a normal Hgb level.[193,194] In addition, Benz et al have recently shown that sleep disorders, common in dialysis patients, improve significantly when Hgb is normalized.[195] Finally, McMahon et al have shown that physical performance of dialysis patients improves significantly when the Hgb is normalized, compared with lower Hgb levels.[196] Although the safety of keeping the Hgb at normal levels needs to be confirmed, particularly in light of the Amgen Normal Hematocrit Cardiac Trial,[158] clinicians must exercise clinical judgment and try to aim for the target Hgb for each patient that will be most beneficial while awaiting the results of additional studies in this area.[197-199]

PUBLIC POLICY ISSUES

There is little debate about the clinical value of treating anemic renal patients with rHuEPO and correcting anemia to the currently recommended target levels. It is clear, however, that this treatment is costly, and the Health Care Financing Administration (HCFA) in the US, the regulatory agency that oversees the Medicare Program, is concerned about the benefits of further expenditures in this area. If one looks only at the change in Hct, and the cost of the rHuEPO used to achieve this change over the past 5 years, it would appear that considerably more rHuEPO is being used, at substantial cost, with only a small improvement in Hct.[200] This should not be surprising, however, because there is not a linear dose-response relationship between dose of rHuEPO and resultant Hct. Two recently published studies on normalizing Hct in hemodialysis patients both showed that a 3- to 4-fold increase in rHuEPO dose is necessary to raise the Hct from the low 30s to the low 40s.[137,158]

A recent study by Collins et al looked not only at the direct cost of rHuEPO, but all Medicare expenditures in a cohort of patients receiving rHuEPO.[201] This comprehensive analysis showed that although the direct costs for rHuEPO rose between 1991 and 1995, total Medicare expenditures were significantly lower for patients with a Hct 33% to 36%. Although part of the explanation for this is likely that patients with higher Hcts are more likely to be healthier and to require less rHuEPO, thus consume fewer health resources overall and cost less, this hypothesis remains to be validated.

At the present time clinicians must provide medical justification if they feel that a patient would benefit from a Hct level maintained above 36.5%. Although the benefits of normal Hcts have not been conclusively proven (see earlier), it is clear that they are real in some patients. HCFA needs to develop a more rational policy for payments for rHuEPO for this group of patients.

The most vexing policy issue involving rHuEPO relates to the current pricing, cost, and reimbursement system. Dialysis facilities negotiate the per unit cost of rHuEPO with the only available supplier in the US, Amgen. Large dialysis chains are able to negotiate substantial discounts, and additional discounts are provided based on hitting certain anemia management targets. Medicare reimburses dialysis facilities $10 for every 1,000 units of rHuEPO administered, whereas the negotiated price is 10% to 30% lower. The result is the increasing reliance of the dialysis facilities on profits from rHuEPO to support the overall operation of the facility. This payment fills in the gap between the cost of providing dialysis services, and the payment per treatment (the composite rate) that has remained static (falling significantly in real dollars) for many years, and no longer is adequate to cover the costs of services. The reliance on profits from rHuEPO (and other pharmaceuticals) in the dialysis setting distorts the marketplace and makes facilities vulnerable to uncontrollable events, like drug price increases, as recently took place with rHuEPO. Independent facilities, those in underserved and rural areas, are the most affected, because they have the least ability to negotiate low prices, and operate close to, if not over the margins. For dialysis to continue to be available as needed, policy makers must reexamine the structure of the entire payment system for outpatient dialysis, including the method of payment of pharmaceuticals such as rHuEPO.

CONCLUSIONS

RHuEPO has improved and prolonged the lives of hundreds of thousands of patients with the anemia of renal failure since its approval in 1989. Since then much has been learned of the physiology and pathophysiology of endogenous EPO as well as the recombinant protein. In the near future Novel Erythropoiesis Stimulating Protein (NESP) will be available to treat renal anemia.[202] NESP is a hyperglycosylated analogue of rHuEPO with a

significantly prolonged half-life in the circulation. Less frequent administration may improve compliance, decrease administration costs, and lead to more stable Hgb levels over time. Clinical trials are currently underway to document its efficacy and safety. Although many advances in the treatment of renal patients are likely in the next decade, it is hard to imagine many that will compare with the impact the rHuEPO has had in just over 10 years.

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